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EFFECT OF REDUCTION OF POTASSIUM ON THE ACTION OF ACETYLCHOLINE ON RABBIT AURICLES

BY

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Spadolini and Domini (1940) were the first to show that minute doses of acetylcholine may stimulate the isolated heart of the guinea-pig and they suggested that this action might be due to a release of adrenaline in cardiac tissue. Hoffmann, Hoffmann, Middleton, and Talesnik (1945) and Haney and Lindgren (1946) have shown that acetylcholine may have a stimulant effect on the heart of the cat or dog after the administration of atropine, and McDowall (1946) has further shown that small doses of acetylcholine may stimulate the cat heart untreated with atropine. Dawes (1946) and de Elío (1947) have shown that acetylcholine reduces the refractory period of the isolated driven auricle of the rabbit, and Wedd and Blair (1946) have demonstrated the direct nature of this effect on nerve-free tissue from the ventricle of the turtle. Recently Burn and Vane (1949) and Bülbring and Burn (1949) have reviewed the literature concerning this subject and as a result of studies of the effect of proguanil and of fatigue on the isolated rabbit auricle have suggested that acetylcholine may play a dual role in the heart. Synthesis of acetylcholine proceeds in the contracting auricle; addition of acetylcholine to produce a concentration above the normal amount present in the tissue causes inhibition of contraction. If the muscle is depressed addition of acetylcholine from without may bring the concentration up to the threshold for normal functioning and bring about stimulation of the heart.

The relationship between the concentration of potassium ions in a tissue and the effects thereon of administered acetylcholine is a close one. In the perfused frog heart increase or decrease of potassium in the perfusing fluid (with appropriate control of the *p*H, saline content, temperature, etc.) reduces the activity of the heart, makes the rhythm irregular and modifies the response to acetylcholine. Similarly increase or decrease of K modifies the contraction of the isolated rabbit auricle.

EXPERIMENTAL

It was decided to test the effect of modification of the ionic content of potassium on the response of the rabbit auricle to added acetylcholine. The beat of the isolated rabbit auricle was recorded as usual at 27° C. in powerfully oxygenated Locke solution with double the normal concentration of glucose. At suitable intervals this fluid was replaced by a modified Locke solution containing half or one and a half the normal amount of potassium. The *p*H was measured and adjusted if necessary by modifying the bicarbonate content to give *p*H 8.2.

The addition of 0.25 μg. acetylcholine in a 75 ml. bath may produce a preliminary stimulation of the auricle of small extent (Fig. 1 *a* and *b*) followed by inhibition. In some specimens this inhibition is not so transient (Fig. 1 *c*) but frequently there is either no effect from addition of such quantities

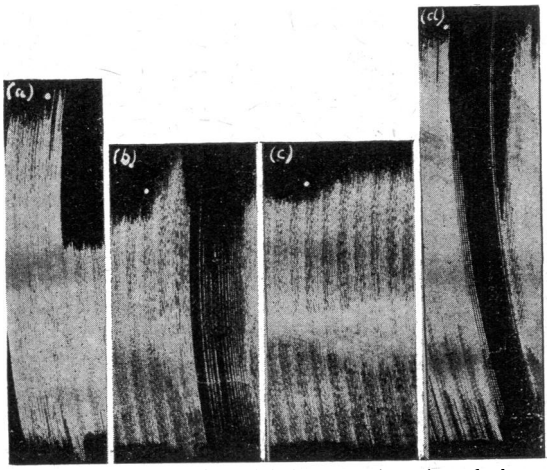

FIG. 1.—Isolated rabbit auricle in normal Locke solution at 27° C. (*a*) and (*b*) show transient stimulation after adding 0.25 μg. acetylcholine to the 75 ml. bath. (*c*) shows a longer stimulation after the same amount of acetylcholine. (*d*) shows inhibition after 1.0 μg.

of acetylcholine or a pure inhibition. The tendency to preliminary transient stimulation develops as the preparation gets older and has been seen in nine out of 47 auricles after two hours beating. In four specimens it was apparent after one hour; it was also seen in two auricles (excluded from the series) which would not beat until they had been treated with 100 μg. acetylcholine followed by washing after a few minutes. Addition of larger quantities of acetylcholine (1 μg. or more) causes inhibition of the activity of these preparations, which may be interrupted by a few beats more powerful than normal (Fig. 1 *d*). Increase of the potassium in the nutrient fluid abolishes this early stimulation with small quantities of acetylcholine, which now have no effect on the heart, or inhibit it.

If the amount of K is reduced the augmentor response to added acetylcholine is enhanced in those preparations which show this effect. Quantities of acetylcholine which, in normal Locke solution, previously or subsequently caused a moderate inhibition of the contraction for a prolonged period might now cause a preliminary stimulation followed by a sharp but transient inhibition, and frequently a prolonged phase of increased activity (Fig. 2 *a*). These effects can be repeated with considerable regularity in those preparations which show the phenomenon (37 out of 45 tests) and are in contrast with the usual action of acetylcholine on the isolated auricle in normal nutrient fluid (Fig. 2 *c* and *d*).

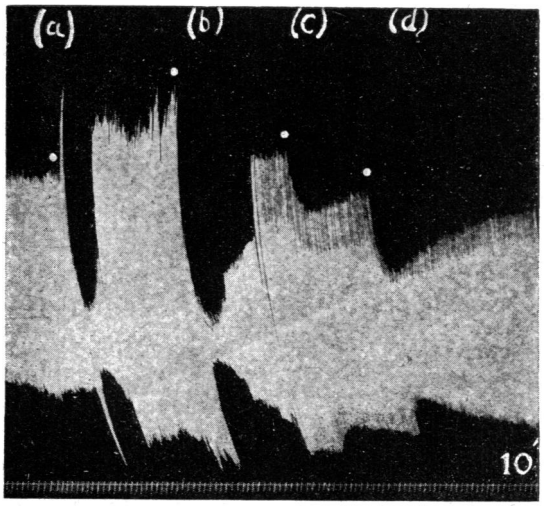

FIG. 2.—Isolated rabbit auricle. (*a*) and (*b*) show the effect of adding 1.0 μg. and 5.0 μg. of acetylcholine respectively, without any wash between, to Locke solution containing half the usual amount of potassium. (*c*) and (*d*) as in (*a*) and (*b*) but in normal Locke solution. For description see text.

DISCUSSION

If isolation of the auricular tissue in Locke solution brings about a state of fatigue and some loss of activity in the muscle, and thus a diminution of synthesis of acetylcholine, addition of a small quantity of acetylcholine will bring the amount of this substance up to the optimal value and cause stimulation of the contraction. This sequence of events is seen in isolated auricles which develop the power of responding to small amounts of added acetylcholine by increased activity, after the preparation has been up for some time. Many specimens never show this property but the same or a similar phenomenon is shown by auricles which have ceased to beat from fatigue and which can be restarted by addition of larger amounts of acetylcholine, with or without washing (Bülbring and Burn, 1949); and by auricles which are inhibited from the beginning, perhaps as a result of rough handling, and which begin to beat after the addition of relatively large doses of acetylcholine, followed by washing out. Reducing the activity of the muscle by increasing the potassium content does not bring about a state of affairs in which acetylcholine causes stimulation. Reduction of the potassium content on the other hand not only modifies the activity of the muscle but also alters the response to acetylcholine, so that amounts of this substance which previously caused inhibition now cause stimulation. Rapid alterations of the response between stimulation and depression may be seen (Fig. 2 *a*) at certain concentration levels of acetylcholine, whereas at higher levels pure depression is observed (Fig. 2 *b*). It may be that an optimum level of potassium is necessary for optimal synthesis of acetylcholine, or for the inhibitory response of the muscle to it. The nature of the response to added acetylcholine would then depend on the condition of the muscle, especially as to its potassium content, and on the amounts of acetylcholine produced in the muscle and applied to it externally, as Burn and Vane suggest. Potassium would appear to play as intimate a role in the positive as in the negative response of cardiac tissue to acetylcholine. According to Jequiér, Plotka, and Petergalvi (1948) the connexion between K and acetylcholine synthesis may lie in the relation between K and degradation of glucose.

SUMMARY

Low concentrations of acetylcholine (3×10^{-9}) may cause a transient stimulation of the isolated rabbit auricle contracting in Locke solution. Higher concentrations inhibit it.

Relative lack of potassium causes doses of acetylcholine, which previously caused a slight transient stimulation, to cause a much larger preliminary stimulation, and some doses which previously inhibited, to stimulate.

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